UNITED STATES PATENT AND TRADEMARK OFFICE

UNITED STATES DEPARTMENT OF COMMERCE
United States Patent and Trademark Office
Address: COMMISSIONER OF PATENTS AND TRADEMARKS
Washington, D.C. 20231
www.uspto.gov

APPLICATION NO.	FILING DATE	FIRST NAMED INVENTOR	ATTORNEY DOCKET NO.	CONFIRMATION NO.
09/491,549	01/26/2000	DAVID CHARLES BAULCOMBE		8682

110 7590 08/28/2002

DANN DORFMAN HERRELL & SKILLMAN
SUITE 720
1601 MARKET STREET
PHILADELPHIA, PA 19103-2307

EXAMINER
PARAS JR, PETER

ART UNIT	PAPER NUMBER
1632	20

DATE MAILED: 08/28/2002

Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No. 09/491,549	Applicant(s) BAULCOMBE ET AL.	
	Examiner Peter Paras	Art Unit 1632	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If the period for reply specified above is less than thirty (30) days, a reply within the statutory minimum of thirty (30) days will be considered timely.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133).
- Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *11 June 2002* .

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1,5-17,21,26-29 and 32-34* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1,5-17,21,26-29 and 32-34* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

11)☐ The proposed drawing correction filed on _____ is: a)☐ approved b)☐ disapproved by the Examiner.

If approved, corrected drawings are required in reply to this Office action.

12)☐ The oath or declaration is objected to by the Examiner.

Priority under 35 U.S.C. §§ 119 and 120

13)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____ .

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

14)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. § 119(e) (to a provisional application).

a) ☐ The translation of the foreign language provisional application has been received.

15)☐ Acknowledgment is made of a claim for domestic priority under 35 U.S.C. §§ 120 and/or 121.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO-1449) Paper No(s) *16* .
4) ☐ Interview Summary (PTO-413) Paper No(s). _____ .
5) ☐ Notice of Informal Patent Application (PTO-152)
6) ☐ Other: .

DETAILED ACTION

Continued Examination Under 37 CFR 1.114

A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicant's submission filed on June 11, 2002 has been entered.

Claims 1, 5-17, 21, 26-29, and 32-34 are pending and are under current consideration.

Priority

Receipt is acknowledged of papers submitted under 35 U.S.C. 119(a)-(d), which papers have been placed of record in the file.

However, upon further review of the instant application, errors, which appear to be inadvertent typographical errors, have been found that do not allow for perfection of foreign priority. In the declaration, power of attorney and power to inspect, received on 4/10/00, under the heading CLAIM UNDER 35 U.S.C. 119, the statement claiming foreign priority benefits refers to 37 U.S.C. 119. The priority statement should have referred to 35 USC 119. In the amendment received on April 23, 2001, Applicants submitted an amendment to page 1 line 2 of the specification that recites a priority claim

under 35 U.S.C. 119(e) to GB application No. 9925459.1 filed October 27, 1999. Priority claims to provisional applications may be filed under 35 U.S.C. 119(e). However, it does not appear that the instant application is claiming the benefit of priority to a provisional application, as a provisional application has not been identified. Although the above errors appear to be inadvertent, priority to GB application No. 9925459.1 cannot be perfected until appropriate amendments correcting the errors have been submitted.

The following are new grounds of rejection under 35 U.S.C. 112, first paragraph:

Claim Rejections - 35 USC § 112, 1st paragraph

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1, 5-17, 21, and 32-34 as amended or originally filed are rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a method of determining the occurrence of target gene silencing or detecting silencing of a target gene in a plant, wherein the presence of short RNA molecules (SRMs), which are 21-25 nucleotides in length and share sequence identity with a target gene is correlative with silencing of a target gene, and wherein the silencing of a target gene is also correlative to a phenotype exhibited by said plant, and wherein the target gene is a transgene or is an endogenous gene that is the same as a transgene; and a method for

isolating SRMs, specific for a target gene, from plants does not reasonably provide enablement for all other methods as claimed. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

The claims are directed to methods of determining the occurrence of target gene silencing in plant comprising obtaining a sample of material from said plant, producing a nucleic acid extract from said sample, and analyzing said extract to determine the presence of short RNA molecules (SRMs), which are 21-25 nucleotides in length, and correlating the presence of said SRMs in the extract with the occurrence of target gene silencing in the plant. The claims are also directed to a method of detecting the silencing of a target gene in a plant comprising the above steps and the additional steps of characterizing SRMs which are present in the extract to determine sequence identity with the target gene, wherein SRMs sharing sequence identity with the target gene are correlative to target gene silencing. The claims are also directed to identifying a silenced target gene by screening a library of genes from an organism and identifying those genes that are similar in identity to any SRMs that have been detected. The claims are also directed to isolating RNA molecules associated with target gene silencing.

The specification discusses that the invention features methods for detecting gene silencing in an organism. See page 3, in line 30 of the specification. The specification discusses that the invention features methods that rely on the detection of small RNA molecules (SRMs), wherein the presence of SRMs is suggestive of gene

silencing. See the specification beginning on page 2 and as consistently discussed throughout. The specification further discusses that the SRMs associated with gene silencing (or otherwise known as post-transcriptional gene silencing [PTGS]) require the presence of exogenous nucleic acid molecules, for example, a transgene or an RNA virus. See the specification on page 2, lines 16-19. While the specification provides extensive teachings pertaining to the isolation and characterization of SRMs related to exogenous nucleic acid molecules in a plant, the specification fails to provide any relevant teachings or specific guidance with regard to the isolation and characterization of SRMs that are not related to exogenous nucleic acid molecules in a plant. Furthermore, the specification fails to even provide any guidance or teachings that demonstrate the existence of SRMs that are not related to exogenous nucleic acid molecules rather only contemplating the existence of such (see for example, page 5, the paragraph bridging to page 6 as well as the first full paragraph on page 6). Thus, as enablement requires the specification to teach how to make and use the claimed invention, the specification fails to enable the methods as claimed. It would have required undue experimentation to practice the methods as claimed without a reasonable expectation of success.

As a first issue, claims 1, 5-7, 21, and 32-34 are directed to a method of determining the occurrence of target gene silencing in a plant. The state of the art suggests that the nature of the invention as it relates to gene silencing, particularly PTGS, is a consequence of the presence of exogenous nucleic acid sequences in a plant. The exogenous nucleic acid sequences may be either transgenes or an RNA

virus; PTGS is thought to be a nucleotide sequence specific natural defense system that targets exogenous nucleic acid molecules by forming sequence specific duplex RNA molecules that are degraded. Evidence for the occurrence of PTGS is thought to be the presence of small RNA molecules, that can be sense or anti-sense, that are about 25 nucleotides in length that are specific for a target nucleic acid molecule. See Hamilton et al (Science, 1999, 286: 950-952) on page 950 columns 1-2. Hamilton et al go on to report that transgenic tomato plants comprising an exogenous copy of ACO cDNA exhibited silencing of the endogenous ACO gene as evidenced by the presence of 25 nucleotide sense and anti-sense ACO RNA molecules. Hamilton et al further report that PTGS can also be observed when a transgene does not have homology to an endogenous gene. For example, transgenic tobacco expressing 35S-β-glucuronidase (GUS) also comprised 25 nucleotide GUS anti-sense RNA molecules suggesting the occurrence of PTGS. See page 950, columns 2-3 bridging to column 1 on page 951. As such the state of the art has set forth that SRM sequences are specific for a particular target sequence. The claims as written however are overbroad in that there is no requirement that the detected SRMs are specific for the target sequence. Further, the working examples provided by the specification do not demonstrate the presence of non-specific SRMs as correlative to target gene silencing; the working examples provide for detection of specific SRMs as a correlation to target gene silencing. See the specification on pages 22-26. Even more, as it has been established in the art that PTGS is also a viral defense mechanism, the mere presence of uncharacterized 25 nucleotide RNA molecules does not guarantee that a target gene has been silenced; a

SRM could be specific for a plant virus. See Hamilton on page 951 column 2. As such, it is unpredictable if the detection of any 25 nucleotide RNA molecules correlates to target gene silencing.

Moreover, there does not appear to be a correlation between the mere presence of SRMs and target gene silencing. In other words since a target gene is silenced, which can be interpreted to mean its expression is diminished or even completely shut off (see the specification, the paragraph bridging pages 9-10), it would seem reasonable that there would be a phenotypic consequence resulting from target gene silencing. This reasoning is supported by Hamilton et al who observe that full length ACO mRNA production is dramatically diminished in transgenic tomatoes that appear to have undergone PTGS. See figure 1, panels A and B which show a direct correlation, in the transgenic tomato lines (T5.2 and T5.3) exhibiting PTGS, between the absence of full length ACO mRNA and the presence of 25 nucleotide RNA ACO molecules. Hamilton et al also report a difference in a more observable phenotype, GPF fluorescence, in a plant exhibiting PTGS, wherein after two to three weeks post-introduction GFP fluorescence was not detectable in *Nicotiana benthamiana*. Since the target gene must be a known gene as taught by the specification (see page 10, beginning on line 20 and bridging to page 11), Applicants have contemplated that the technology embraced by the instant invention can be used to silence any gene deemed appropriate in order to eliminate or reduce unwanted traits. Such contemplations are interpreted to clearly read on a phenotypic consequence as a result of gene silencing. The claims do not require any phenotype in a plant as a correlation to target gene silencing in conjunction

with detection of SRMs. In the absence of a phenotype the skilled artisan would not understand that a gene is silenced in a plant and would not attempt to detect SRMs. In the absence of a phenotype it is unpredictable if a target gene is silenced. The evidence of record has not provided guidance or relevant teachings that would enable the skilled artisan to practice the invention as claimed in the absence of a correlation between a phenotype and gene silencing. Given the nature of the invention and the lack of guidance provided by the evidence of record it would have required undue experimentation for one of skill in the art to practice the invention as claimed without a reasonable expectation of success.

As a second issue, claims 8-10 are directed to a method of detecting the silencing of a target gene, wherein any SRMs that are detected, which have sequence similarity to the target gene are correlated to silencing of the target gene. The issue with these claims is whether the detection of sequence specific SRMs is sufficient to imply that a target gene is silenced. As discussed above the specification has contemplated that silencing a target gene can change a plant's phenotype (see page 10, beginning on line 20 and bridging to page 11). It is known that the members of gene families can comprise overlapping regions of sequence homology so the fact that a SRM has sequence identity or similarity with a target gene may not be a sufficient correlation to silencing of that particular target gene; the SRMs could be specific for other genes, having different functions, in a gene family. A correlation between a phenotype and detection of SRMs would seem appropriate, particularly since the target gene is known, and more particularly because a change in phenotype is a clear

indication that a gene is silenced. Without a change in phenotype as an indication of target gene silencing, it is unknown why a skilled artisan would believe that a target gene has been silenced and attempt to detect SRMs that may be associated with silencing. Without a phenotypic change it is unpredictable if a target gene has been silenced and the skilled artisan would not know how to practice the method as claimed, particularly because the instant specification has taught that it may be desirable to silence genes conferring unwanted traits in a plant.

As a third issue, claim 11 is directed to identifying a silenced target gene in a plant comprising screening a library of genes for sequence identity to SRMs. As discussed in the specification on page 10, beginning on line 20, a target gene must be a known gene unless otherwise qualified. The steps of method as claimed appear to be directed to identifying unknown genes that may be silenced based on sequence identity. Such method steps are contemplated in the specification for identifying unknown targets. See the specification on pages 5-6. If a target gene is known, then there does not seem to be any point in identifying it again. If the target gene is unknown, then the instant specification has not provided adequate guidance for practicing the claim as written. Guidance for practicing the invention as claimed is lacking because the specification has contemplated that PTGS may underlie plant developmental processes that would allow plant (unknown) genes to be targeted naturally to satisfy a developmental program. The state of the art at the time of filing as evidenced by Hamilton et al clearly suggests that PTGS results from the introduction of exogenous nucleic acid molecules in plants. Since the nature of PTGS appears to be related to

introduction of exogenous nucleic acid it is unpredictable if PTGS occurs naturally within a plant during development. Moreover, in light of such it is unpredictable if any 25 nucleotide RNA molecules in the absence of exogenous nucleic acid are correlatable to target gene silencing, so any 25 nucleotide RNA fragment could be used to detect naturally silenced target genes. The working examples provided by the instant specification only relate to the introduction of exogenous nucleic acid molecules to trigger PTGS. Additionally, the claim requires that the library in step (vii) is obtained from an organism while the preamble requires the identification of a silenced target gene in a plant. The term organism is extremely broad not only encompassing plants but all animals as well. It would be unpredictable if screening a gene library from any organism would allow the identification of a silenced target gene in a plant. The specification has not provided any relevant teachings or guidance that correlates screening of a gene library obtained from any organism with a SRM obtained from a plant to identify a silenced gene in a plant. In light of the preceding it would have required undue experimentation for one of skill in the art to practice the invention as claimed.

As a fourth issue, claims 12-16 are directed to a process for identifying one or more RNA molecules associated with target gene silencing from a sample of material by obtaining RNA molecules from an extract of material. As taught by Hamilton et al PTGS is a sequence specific phenomenon that occurs as a result of the introduction of exogenous nucleic acid molecules in a plant. See Hamilton on page 950 in column 1 as well as the abstract. The claims however do require any specificity of the purified RNA

molecules to a silenced target gene. In fact step (b) of claim 12 allows for the isolation of any RNA molecule. It is unpredictable if any isolated RNA molecule is associated with gene silencing. The state of the art as evidenced by Hamilton and supported by the instant specification suggests that the RNA molecules associated with target gene silencing in plants are of the 25 nucleotide variety, with a sequence specificity to the target gene. Furthermore, SRMs are known to exist in nature for purposes other than silencing a target gene. Hamilton has discussed that PTGS occurs in response to viral mRNAs in plants, resulting in the production of SRMs specific for a plant virus. The instant specification has not provided any guidance or relevant teachings that suggest that any RNA molecule can be associated with target gene silencing. Moreover, claims 12-15 are not directed to SRMs, let alone an RNA molecule with sequence specificity for a target gene. Claim 16 while directed to SRMs, does not indicate that the SRM is specific for a target gene. Finally, all the claims (12-16) broadly encompass a sample of any material. While, the specification supports the existence of SRMs in plants and *C. elegans* there does not appear to be any evidence provided for the existence of SRMs in a sample of any material. Applicants are reminded that the elected subject matter under examination is limited to plants. The state of the art also corroborates the existence of SRMs in plants for example. See Hamilton throughout the entire document. As such it is unpredictable if any RNA molecule, particularly a SRM, could be isolated from any sample of material, particularly when the molecule is required to be associated with target gene silencing. Given the lack of guidance provided by the

instant specification it would have required undue experimentation to practice the invention as claimed.

As a fifth issue, claim 17 is directed to isolating SRMs for a target gene from any organism. While, the specification supports the existence of SRMs in plants and *C. elegans* there does not appear to be any evidence provided for the existence of SRMs in a sample obtained from any organism. Applicants are reminded that the elected subject matter under examination is limited to plants. The state of the art also corroborates the existence of SRMs in plants for example. See Hamilton throughout the entire document. As such it is an unpredictable and undeveloped aspect of the art if SRMs associated with target gene silencing exist in organisms other than plants and *C. elegans.*

Given the unpredictability and undeveloped state of the art it would have required undue experimentation to practice the invention as claimed.

Claims 26-29 as originally filed or amended are rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

The claims are directed to a DNA construct comprising a promoter operably linked to DNA that encodes a SRM or an anti-sense RNA molecule capable of targeting a region of a target gene. The claims are also directed to a host cell containing the

same construct and a plant comprising the same host cell. The claims are further directed to a method of silencing a target gene in a plant by using the same construct to create a transgenic plant.

Vas-Cath Inc. v. Mahurkar, 19USPQ2d 1111 (Fed. Cir. 1991), clearly states that "applicant must convey with reasonable clarity to those skilled in the art that, as of the filing date sought, he or she was in possession *of the invention*. The invention is, for purposes of the 'written description' inquiry, *whatever is now claimed.*" Vas-Cath Inc. v. Mahurkar, 19USPQ2d at 1117. The specification does not "clearly allow persons of ordinary skill in the art to recognize that [he or she] invented what is claimed." Vas-Cath Inc. v. Mahurkar, 19USPQ2d at 1116.

The DNA sequences that encode all short RNA molecules or anti-sense RNA molecules encompassed within the genus of SRMs have not been disclosed. Based upon the prior art there is expected to be sequence variation among the species of cDNA, which encode SRMs as SRMs are thought to be specific for target genes. The specification discloses that SRMs, either in sense or anti-sense orientation, are generated in response to the presence of exogenous nucleic acid molecules in a plant. The specification however has not disclosed the sequences of any of the DNA molecules, which encode SRMs, to be used in the claimed DNA construct. There is no evidence on the record of a relationship between the structures of the DNA molecules encoding any SRMs that would provide any reliable information about the structure of DNA molecules within the genus. There is no evidence on the record that SRMs had known structural relationships to each other; the art indicated that there is variation

between DNA sequences encoding SRMs, as they are specific for different target genes. The claimed invention as a whole is not adequately described if the claims require essential or critical elements which are not adequately described in the specification and which is not conventional in the art as of applicants effective filing date. Possession may be shown by actual reduction to practice, clear depiction of the invention in a detailed drawing, or by describing the invention with sufficient relevant identifying characteristics such that a person skilled in the art would recognize that the inventor had possession of the claimed invention. Pfaff v. Wells Electronics, Inc., 48 USPQ2d 1641, 1646 (1998).

In the instant case the claimed embodiments of DNA molecules that encode SRMs or anti-sense RNA molecules encompassed within the genus of SRMs lack a written description. The specification fails to describe what DNA molecules fall into this genus and it was unknown as of Applicant's effective filing date that any of these DNA molecules would have the property of encoding a SRM that can silence a target gene. The skilled artisan cannot envision the detailed chemical structure of the encompassed DNA molecules, and therefore conception is not achieved until reduction to practice has occurred, regardless of the complexity or simplicity of the method of isolation. Adequate written description requires more than a mere statement that it is part of the invention and reference to a potential method of isolating it. See *Fiers v. Revel*, 25 USPQ2d 1601, 1606 (Fed. Cir. 1993) and *Amgen Inc. v. Chugai Pharmaceutical Co. Ltd.*, 18 USPQ2d 1016 (Fed. Cir. 1991).

One cannot describe what one has not conceived. See *Fiddes v. Baird*, 30 USPQ2d 1481, 1483. In *Fiddes*, claims directed to mammalian FGF's were found to be unpatentable due to lack of written description for that broad class. The specification provided only the bovine sequence.

In view of the above considerations one of skill in the art would not recognize that applicant was in possession of the necessary common features or attributes possessed by member of the genus of DNA fragments encoding SRMs. Moreover, the art has recognized that there would be variation among the species of the genus of DNA molecules that encode SRMs as SRMs appear to be specific for particular target genes. Therefore, Applicant was not in possession of the genus of DNA molecules that encode SRMs as encompassed by the claims. University of California v. Eli Lilly and Co., 43 USPQ2d 1398, 1404, 1405 held that to fulfill the written description requirement, a patent specification must describe an invention and do so in sufficient detail that one skilled in the art can clearly conclude that "the inventor invented the claimed invention."

Claim Rejections - 35 USC § 112, 2nd paragraph

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 11-17 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 11 recites the limitations "said organism" and "the organism" in steps (vii) and (viii) respectively. There is insufficient antecedent basis for this limitation in the claim.

Claim 12 is incomplete as written. The claim is directed to a process for isolating one or more RNA molecules associated with target gene silencing from a sample of material. The claim is incomplete because the steps of the method do not positively relate back to the preamble of the claim. Claims 13-16 depend from claim 12. It is noted that claim 17 while depending from claim 12 is not incomplete as written as the steps of claim 17 relate back to isolating SRMs as recited in the preamble.

Claim 17 recites the limitations "said organism" and "said organism" in steps (i) and (ii) respectively. There is insufficient antecedent basis for this limitation in the claim.

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (a) the invention was known or used by others in this country, or patented or described in a printed publication in this or a foreign country, before the invention thereof by the applicant for a patent.
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

The previous rejection of claims 1, 5-10, 12-17, 21, 26-29 and 32-34 under 35 U.S.C. 102(a) as being anticipated by the Leysin Meeting Presentation (IDS doc. C6) is

withdrawn in view of Applicants declaration filed on June 11, 2002, which states that the subject matter presented in Leysin was the result of a collaborative inventive effort by both inventors, Baulcombe and Hamilton.

The following are new grounds of rejection under 35 U.S.C. 102:

Claims 12-14 are rejected under 35 U.S.C. 102(b) as being anticipated by Cleuziat et al (US 5,849,547).

The claims are directed to a process for isolating one or more RNA molecules associated with target gene silencing from a sample of material comprising producing a nucleic acid extract from the sample, purifying the extract to produce purified RNA molecules. The claims are further directed to separating the purified RNA molecules on a 15% polyacrylamide gel, containing 7M urea, which uses a running buffer of 0.5x TBE. The claims are also directed to transferring the RNA molecules from the gel to a membrane using current. It would appear that the claims as a whole broadly embrace the process of Northern blotting.

It is noted that the preamble is directed to isolating one or more RNA molecules associated with target gene silencing. When relying on the specification for a working definition of what are the RNA molecules associated with target gene silencing the Examiner's attention is directed to short RNA molecules (approximately 25 nucleotides in length), which appear to be the RNA molecules associated with target gene silencing. However, the steps of the method are only directed to isolation of total RNA and would not provide for isolation of one or more RNA molecules associated with target gene

silencing from other RNA molecules. Because the method steps would not allow for the isolation of RNA molecules associated with target gene silencing the preamble is not given considerable weight for the purposes of the instant rejection.

Cleuziat et al teaches that the target nucleic acid molecule (i.e. the nucleic acid molecule of interest) can be RNA or DNA, wherein the target nucleic acid molecule can be extracted from a biological sample. See column 5 beginning in line 66 and bridging to column 6 through line 7. Cleuziat et al further teach that nucleic acids may be purified from a biological extract by phenol chloroform extraction, chromatography, ion exchange, electrophoresis, etc... See column 18 in lines 8-19. Cleuziat teaches that the purified target nucleic acid molecule can be amplified and then analyzed by electrophoresis on a denaturing polyacrylamide gel, particularly a 15% acrylamide gel comprising 7M urea. See column 27, lines 19-39. The running buffer typically used in the top reservoir of the electrophoresis is 0.5x TBE. Although Cleuziat does not teach the use of 0.5x TBE, the use of such is standard in molecular biology protocols and is evidenced by Molecular Cloning (1989), which is a laboratory handbook of molecular biology protocols. See Molecular Cloning on page 13.54. Cleuziat et al goes on to teach that the molecules separated on the 15% polyacrylamide gel can then be transferred to a nylon membrane using a "mini trans-blot electrophoretic transfer cell" apparatus under an electric field of 35 volts/hour/cm. See column 27, lines 40-48. Thus, the teachings of Cleuziat et al anticipate all of the instant claim limitations.

Claim 12 is rejected under 35 U.S.C. 102(b) as being anticipated by Mueller et al (1995, The Plant Journal, 7(6): 1001-1013).

Claim 12 is directed to a process for isolating one or more RNA molecules associated with target gene silencing from a sample of material comprising producing a nucleic acid extract from the sample, purifying the extract to produce purified RNA molecules. The claim broadly encompasses a method of RNA isolation.

It is noted that the preamble is directed to isolating one or more RNA molecules associated with target gene silencing. When relying on the specification for a working definition of what are the RNA molecules associated with target gene silencing the Examiner's attention is directed to short RNA molecules (approximately 25 nucleotides in length), which appear to be the RNA molecules associated with target gene silencing. However, the steps of the method are only directed to isolation of total RNA and would not provide for isolation of one or more RNA molecules associated with target gene silencing from other RNA molecules. Because the method steps would not allow for the isolation of RNA molecules associated with target gene silencing the preamble is not given considerable weight for the purposes of the instant rejection.

Mueller et al teach a method of extracting RNA from plant leaves. See page 1011, in the section entitled "Experimental Procedures", column 2 under the heading "Extraction of total RNA and DNA", the whole paragraph. Mueller et al teaches the homogenization of leaves in a lysis buffer and separation of RNA by LiCl precipitation. Thus, the teachings of Mueller et al anticipate all of the instant claim limitations.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The previous rejection of claims 1, 5-17, 21, 26-29, and 32-34 under 35 U.S.C. 103(a) as being unpatentable over Waterhouse et al and Wasseneger et al taken with Doughtery et al is withdrawn in view of Applicants arguments filed on June 11, 2002. Particularly, Applicants have argued that Doughtery et al, as relied on by the Examiner, discloses that 21nt SRMs were detected in *C. elegans* and that the instant claims are directed to plants only. As such Applicants have argued that there would be no apparent motivation to extrapolate the *C. elegans* data to plants, in order to detect 21 nt SRMs in plants. The Examiner has found this argument persuasive. In light of such there would have been no apparent motivation to look specifically for 21-25nt SRMs in plants, although the prior art has suggested that SRMs from 10-100nt may exist in plants.

The following are new grounds of rejection under 35 U.S.C. 103(a):

Claims 12-15 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cleuziat et al (US 5,849,547).

The claims are directed to a process for isolating one or more RNA molecules associated with target gene silencing from a sample of material comprising producing a nucleic acid extract from the sample, purifying the extract to produce purified RNA molecules. The claims are further directed to separating the purified RNA molecules on a 15% polyacrylamide gel, containing 7M urea, which uses a running buffer of 0.5x TBE. The claims are also directed to transferring the RNA molecules from the gel to a membrane using current and labeling the transferred molecules with a radioactive probe. It would appear that the claims as a whole broadly embrace the process of Northern blotting.

Cleuziat et al teaches that the target nucleic acid molecule (i.e. the nucleic acid molecule of interest) can be RNA or DNA, wherein the target nucleic acid molecule can be extracted from a biological sample. See column 5 beginning in line 66 and bridging to column 6 through line 7. Cleuziat et al further teach that nucleic acids may be purified from a biological extract by phenol chloroform extraction, chromatography, ion exchange, electrophoresis, etc... See column 18 in lines 8-19. Cleuziat teaches that the purified target nucleic acid molecule can be amplified and then analyzed by electrophoresis on a denaturing polyacrylamide gel, particularly a 15% acrylamide gel comprising 7M urea. See column 27, lines 19-39. The running buffer typically used in the top reservoir of the electrophoresis is 0.5x TBE. Although Cleuziat does not teach the use of 0.5x TBE, the use of such is standard in molecular biology protocols and is evidenced by Molecular Cloning (1989), which is a laboratory handbook of molecular biology protocols. See Molecular Cloning on page 13.54. Cleuziat et al goes on to

teach that the molecules separated on the 15% polyacrylamide gel can then be transferred to a nylon membrane using a "mini trans-blot electrophoretic transfer cell" apparatus under an electric field of 35 volts/hour/cm. See column 27, lines 40-48.

Cleuziat et al differs from the claimed invention by teaching labeling of RNA molecules on the hybridization membrane with a non-radioactive probe.

However at the time the claimed invention was made, it was well within the purview of the ordinarily skilled artisan to use radioactive probes for labeling nucleic acid molecules on a hybridization membrane, particularly RNA molecules. This is evidenced by Cleuziat et al in the description of the related arts in column 4, lines 39-62, who discusses that a variety of detection methods can be used for labeling nucleic acid molecules including RNA, which may comprise using radioactive and non-radioactive probes. Since Cleuziat et al discuss that use of radioactive probes is well known within the state of the art, it would appear that use of a radioactive probe is quite common and is an obvious variation, and not a patentable limitation on its own, that could be practiced in the common molecular biology technique of Northern blotting, which is what the instant claims appear to encompass.

Accordingly, in view of the routine state of the art as represented by Cleuziat et al, it would have been obvious to use a radioactive probe to label RNA molecules on a hybridization membrane. One of ordinary skill in the art would have been sufficiently motivated to use a radioactive probe for labeling RNA molecules as it is an obvious variation in routine molecular biology methods as evidenced by Cleuziat et al.

Thus, the claimed invention, as a whole, was clearly prima facie obvious in the absence of evidence to the contrary.

Conclusion

No claim is allowed. Claims 1, 5-11, 16-17, 21, 26-29, and 32-34 appear to be free of the prior art of record but are subject to other rejections. The point of novelty of the instant application appears to be the fact that the SRMs are 21-25 nucleotides as recited in claim 1 for example. However, it is also important that the SRMs are sequence specific for a target gene and so that the presence of such is correlative with silencing of a target gene, and wherein the silencing of a target gene is also correlative to a phenotype exhibited by said plant, and wherein the target gene is a transgene or is an endogenous gene that is the same as a transgene. Amending the claims, using acceptable language, to read on the preceding limitations may be sufficient to overcome the remaining rejections of record.

Any inquiry concerning this communication or earlier communications from the examiner(s) should be directed to Peter Paras, Jr., whose telephone number is 703-308-8340. The examiner can normally be reached Monday-Friday from 8:30 to 4:30 (Eastern time).

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Deborah Reynolds, can be reached at 703-305-4051. Papers related to this application may be submitted by facsimile transmission. Papers should be faxed via the PTO Fax Center located in Crystal Mall 1. The faxing of such papers must conform with the notice published in the Official Gazette, 1096 OG 30 (November 15, 1989). The CM1 Fax Center numbers are (703) 308-4242 and (703) 305-3014.

Inquiries of a general nature or relating to the status of the application should be directed to Patsy Zimmerman whose telephone number is (703) 308-0009.

Peter Paras, Jr.
Art Unit 1632

Peter Paras
Art Unit 1632